

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1. (presently amended) A silk-fiber-based matrix ~~having a wire-rope geometry for use in producing a predetermined type of ligament or tendon ex vivo~~ composition comprising sericin-extracted silkworm fibroin fibers, said fibers being biocompatible and helically organized, wherein said matrix supports ingrowth of cells around said fibroin fibers and is biodegradable.
2. (originally filed) The matrix as recited in claim 1, wherein said silk-fiber-based matrix is comprised of silk fibroin selected from the group consisting of silks from silkworms, ~~silks from spiders~~, silks from genetically engineered cells, transgenic plants and animals, silks from cultured cells, native silk, silk from cloned full or partial sequences of native silk genes, and silk from synthetic genes encoding silk or silk-like sequences.
3. (originally filed) The matrix as recited in claim 2, wherein the silk-fiber based matrix comprises fibroin obtained from *Bombyx mori* silkworm fibers.
4. (originally filed) The matrix as recited in claim 1, wherein the matrix comprises a composite of silk and collagen fibers.
5. (originally filed) The matrix as recited in claim 1, wherein the matrix comprises a composite of ~~silk and~~ silk fibroin fibers and one or more silk foams, films, meshes or sponges.
6. (originally filed) The matrix as recited in claim 1, wherein the matrix comprises a composite of silk and one or more degradable polymers selected from the group consisting of Collagens, Polylactic acid or its copolymers, Polyglycolic acid or its copolymers, Polyanhydrides, Elastin, Glycosamino glycans, and Polysaccharides.

7. (canceled)
8. (originally filed) The matrix as recited in claim 1, further comprising pluripotent or fibroblast cells seeded on said matrix.
9. (presently amended) The matrix as recited in claim 9 8, wherein said pluripotent or fibroblast cells are autologous.
10. (presently amended) The matrix as recited in claim 9, wherein said pluripotent or fibroblast cells are ~~allogenic~~ allogeneic.
11. (originally filed) The matrix as recited in claim 9, wherein said pluripotent cells are selected from the group consisting of bone marrow stromal cells and adult or embryonic stem cells
12. (originally filed) The matrix as recited in claim 9, wherein said fibroblast cells are mature human ACL fibroblast cells.
13. (originally filed) The matrix as recited in claim 9, wherein said pluripotent or fibroblast cells proliferate and differentiate on said matrix to form said predetermined ligament or tendon.
14. (originally filed) The matrix as recited in claim 9, further comprising a surface modification agent which enhances ~~poliferation~~ proliferation and differentiation of said pluripotent or fibroblast cells on said matrix.
15. (presently amended) The matrix as recited in claim 1, wherein said matrix comprises a shape of a ligament or tendon ~~produced~~ is selected from the group consisting of anterior cruciate ligament, posterior cruciate ligament, rotator cuff tendon, medial collateral ligament of the elbow, flexor tendon of the hand, ligaments and tendons of the temporomandibular joint, and lateral ligament of the ankle.

16. (presently amended) The matrix as recited in claim 15, wherein said ligament ~~produced~~ is an anterior cruciate ligament.

17. – 173. (canceled)

174. (new) A silk-fiber-based matrix comprising helically organized sericin-extracted silk fibers and bone marrow stromal cells attached thereto.

175 (new) A composition consisting essentially of sericin-extracted silk fibroin fibers, said fibers being biodegradable and organized into a matrix comprising an ultimate tensile strength of greater than 2000N and a linear stiffness of between 100-600N/mm, wherein sericin is completely removed from said fiber.

176 (new) The matrix of claim 14, wherein said surface modification comprises an arginine-glycine-aspartate peptide.

177. (new) The matrix of claim 1, wherein said helically-organized fibers comprise a cylindrical shape.

178 (new) The matrix of claim 1, wherein said matrix comprises an ultimate tensile strength of greater than 2000N and a linear stiffness of between 100-600N/mm.

REMARKS

The claimed invention is drawn to a sericin-purified silk fibroin fiber based matrix with mechanical properties, which have not been previously achieved using extracted silk fibers.

Silk, as the term is generally known in the art, means a filamentous product secreted by an organism such as a silkworm or spider. Fibroin is produced and secreted by a silkworm's two silk glands. As fibroin leaves the glands, it is coated with sericin, a glue-like substance.

Two types of silk are generally known in the art: (1) naturally-occurring silk, and (2) silk suture. Naturally-occurring silk (i.e., sericin-coated fibroin fibers) is non-biocompatible. Moreover, sericin in silk is antigenic and is associated with an adverse tissue reaction. However, removal of sericin from silk changes the ultrastructure of the fibroin fibers, results in loss of mechanical strength, and leads to a fragile structure. Applicants have developed methods of removing sericin from silk in a manner that preserves the mechanical strength of the fibers and have further organized sericin-free silk fibroin fibers into defined geometric configurations, which confer tensile strength which has not previous been achieved with sericin-extracted silk fibroin fibers. Conventional suture silk is coated with a wax-like substance to prevent cell ingrowth and is non-biodegradable.

Claims 1-16 and 174-175 are pending. Claims 17-173 have been canceled. Claims 1, 9, 10, 15 and 16 were amended. New claims 174-178 were added. The amendment to claim 1 and new claims 174-175 are supported by disclosure at page 5, lines 8-10; page 2, line 3; page 6, lines 25-29 (as well as Figs. 1A-C); page 8, lines 16-23 (as well as Fig. 9) and, page 14, line 7; of the specification. New claims 175 and 178 are further supported by disclosure at page 11, lines 3-30; page 7, lines 19-25 (as well as Figs. 4A-D). New claim 176 is supported by disclosure at page 13, lines 19-20, of the specification. New claim 177 is supported by disclosure at page 14, line 9, of the specification.

APPLICANTS: Altman et al.
SERIAL NUMBER: 10/008,924

No new matter has been added by this amendment.

35 U.S.C. § 112, second paragraph

Claims 1-80, 128-137 and 174 were rejected for indefiniteness. With respect to the claim term “wire rope geometry”, the Examiner stated:

The claims are confusing and unclear by reciting “wire rope geometry” which is uncertain as to meaning and scope, and is unclear as to structure required. Such language does not appear to be art recognized for defining a particular structure.

The claims have been amended to delete the term “wire rope geometry”. Claim 1 now requires that the silk fibers be helically organized. Applicants submit that the term helical organization defines a well-understood structure or configuration. Claim 15 further requires that the matrix be in the shape of an anterior cruciate ligament, posterior cruciate ligament, rotator cuff tendon, medial collateral ligament of the elbow, flexor tendon of the hand, ligaments and tendons of the temporomandibular joint, and lateral ligament of the ankle. The shape of these anatomical structures is also well known in the art.

Applicants therefore submit that the meaning and scope amended claims are clear and respectfully request withdrawal of this rejection.

35 U.S.C. § 103

Li et al. in view of Lewis et al. and Takezawa et al.

Claims 1-3, 5, 8-19, 21, 24-36, 38, 41-56, 58, 61-71, 73, 76-80, 128, 129, 133-137, and 174 were rejected for obviousness over Li et al. in view of Lewis et al. and Takezawa et al. The Examiner states:

It would have been obvious to prepare the filamentous matrix of Li et al. from silk in view of Lewis et al. suggesting preparing a matrix for tissue reconstruction

from silk and Takezawa et al. using a silk mesh as a culture carrier. The filamentous matrix of Li et al. would inherently have a wire rope geometry and be capable of use to produce ligament or tendon tissue in vitro. Seeding the matrix with specific cells would have been a matter of obvious choice depending on the type of tissue being produced. Forming bioengineered tissue using the matrix would have been obvious since this is a well known use of such a matrix.

The claims have been amended to require a biodegradable silk matrix with a helical organization made from biocompatible silk fibroin fibers from which sericin has been removed. Li et al. describe a scaffold or filamentous cell-supporting matrix of a variety of non-biodegradable, biocompatible materials. Among the listed non-biodegradable materials is silk. As is described in the specification and shown in Fig. 1a of the present application, native silk contains sericin, a glue-like coating. The composition of the claims is distinguished from that described by Li et al., because the claims require a sericin-free silkworm fibroin fiber that is biodegradable, while Li et al. describe only an unprocessed sericin-coated silk material that is nonbiodegradable.

Lewis et al. describe spider silk and suggest a matrix material for reconstruction of bone and connective tissue, but fail to describe a matrix containing a seracin-extracted silkworm fibroin fibers with the geometric configuration required by the amended claim.

Takezawa et al. describe cell culture carrier, i.e., a vessel in which to grow cells, which contains a mesh of silk suture material, which is non-biodegradable. In contrast, the claims require biodegradable matrix containing a seracin-extracted silk, i.e., silk that has been processed to remove sericin.

None of the cited references describes or suggests a sericin-free, biocompatible, biodegradable silkworm fibroin fiber matrix required by the amended claims. Therefore, the

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amended claims are non-obvious over the combination of Li et al. in view of Lewis et al. and Takezawa et al.

Li et al. in view of Lewis et al. and Takezawa et al. in further view of Mansmann et al.

Claims 4, 6, 20, 22, 37, 39, 57, 59, 72, 74, 130, and 131 were rejected for obviousness over Li et al. in view of Lewis et al. and Takezawa et al. in further view of Mansmann et al. The recited subset of claims recites a combination of sericin-free silk and another degradable polymer such as collagen. Mansmann et al. was cited for a description of a cell scaffold of collagen fibers. The Examiner stated:

It would have been obvious to use collagen fibers in combination with the filamentous silk matrix of Li et al. or use collagen to form a composite with the silk of Li et al. in view of Mansmann disclosing preparing a matrix from collagen fibers and Li et al. suggesting coating the silk matrix with collagen (col. 2, lines 59).

As is discussed above, none of the cited references describes or suggests a sericin-free, biocompatible, biodegradable silkworm fibroin fiber matrix. Mansmann et al. fail to provide disclosure for elements of the amended claims that missing in Li et al. in view of Lewis et al. and Takezawa et al. Li et al. simply describes “silk”, i.e., fibroin with a sericin coating, a fiber that is non-biocompatible and non-biodegradable. Further coating such a composition with collagen does not result in or suggest the compositions defined by the amended claims.

Li et al. in view of Lewis et al. and Takezawa et al. in further view of Shalaby et al.

Claims 7, 23, 40, 60, 75, and 132 were rejected for obviousness over Li et al. in view of Lewis et al. and Takezawa et al. in further view of Shalaby et al. Claims 7, 23, 40, 60, 75, and 132 have been canceled. Therefore, this rejection is moot.